CLINICAL SYNDROMES ASSOCIATED WITH FUNGAL INFECTIONS

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I. Fever in Compromised Host

A. Disseminated granulomatous infections

The first case is a 41 year old white male from Seagoville, Texas, who presented to the DVAH in the fall of 1975, with persistent fever, chills, and weight loss. He had presented initially to the DVAH in May 1974, with hepatosplenomegaly and a low WBC count with predominantly atypical lymphocytes. A bone marrow aspirate and biopsy revealed cells consistent with leukemic reticuloendotheliosis (hairy cell leukemia). A splenectomy was performed but he continued to have frequent bacterial infections following a stormy postoperative course, including staphylococcal bacteremia, pseudomonas bacteremia, secondary to an infected intravenous catheter site, and E. coli pneumonia. In early 1975, he was admitted with fever and chills, abnormal liver function studies, but no pulmonary infiltrate. Cultures of both sputum and liver biopsy tissue grew Mycobacterium kansasii. He was begun on isoniazid, ethambutol and rifampin and he improved but he began to spike temperatures again when he developed an erythema multiforme illness with synovitis. The isoniazid was stopped with subsequent improvement in the synovitis; however, the dermatologists diagnosed the skin eruption as Sweet's syndrome (acute febrile neutrophilic dermatosis), since a biopsy showed polymorphonuclear leucocytes (1). He eventually improved, leaving the hospital afebrile after two months on ethambutol, rifampin and streptomycin (twice a week). He gained weight and did well.

In October he developed fever again. His chest x-ray revealed no pulmonary infiltrate, liver function studies were normal, and there was no change in his blood status. Liver scan was normal, a bone marrow biopsy showed no granuloma, although leukemic cells were present. Blood, bone marrow, CSF and sputum for acid fast, fungi and routine organisms were negative (Table 1). Serological tests (fungal, bacterial agglutinins, cytomegalovirus and Toxoplasma titers) were negative. Patient was discharged on two additional antituberculous drugs with no subsequent change in his course. When he continued to have fever daily, he was readmitted in early December. A gallium scan and IVP were negative, chest x-ray showed no changes and the only new physical finding was a left upper eyelid lesion, which grew Staphylococcus epidermidis but failed to respond to cloxacillin and local care. Blood and sputum cultures were repeated. Liver biopsy showed leukemic infiltration but no granuloma. Although his fever continued, he was discharged on antituberculous medication, after the drugs had been stopped for at least a week to rule out drug fever. He was readmitted on New Year's Eve, 1975, because he was quite toxic with fever up to 104° to 105° daily. This time blood cultures which had been held for 3 weeks and the liver biopsy culture showed fungi consistent with Histoplasma capsulatum. Bone marrow showed one monocyte containing Histoplasma organisms. Cultures of bone marrow and the left upper eyelid also grew Histoplasma, but pharyngeal and sputum cultures were negative. The histoplasma yeast C.F. titer from early December showed 1:32. (Table 1).

TABLE 1

DATA: DISSEMINATED HISTOPLASMOSIS

	Date		
	10/75	12/76	1/76
MYCOLOGICAL DATA			
Bone marrow	Neg	-	Pos
Blood	Neg	Pos	Pos
Liver	-	Pos	-
Sputum	Neg	Neg	Neg
IMMUNOLOGICAL DATA			
Yeast	Neg	1:32	1:64
Histoplasmin	Neg	Neg	Neg

The patient was begun on 5 mg amphotericin intravenously and advanced slowly to 45 mgs daily. The patient tolerated the drug quite well; in fact, he gained weight, became afebrile and began to feel considerably better. His antituberculous drugs were continued, although all the cultures submitted for acid fast organisms were negative. Three weeks into the course of amphotericin, he developed an erythematous, pruritic eruption over the entire body and all medication was discontinued. Before amphotericin could be reinstituted, he developed a right upper lobe pneumonia for which he received carbenicillin and gentamicin. White count at this time showed 1800 with 79% atypical lymphocytes. Blood cultures failed to grow any pathogen so he was treated for 8 days. The drugs were discontinued when he developed a drug eruption. Amphotericin was then reinstituted when the rash cleared, since it was felt he had not received an adequate course as yet. He continued to have temperature and repeat blood cultures revealed a rapid-growing Mycobacteria species (Group IV , not M. fortuitum) which was sensitive only to kanamycin. He was begun on kanamycin. The patient failed to regain strength and in early March 1976, he developed a new pulmonary infiltrate. A transtracheal aspirate revealed small bipolar-staining gram negative rods in pairs with a capsule. He was begun on cefazolin, chloramphenicol, tobramycin and carbenicillin but he died the next day. The organism subsequently was shown to be Klebsiella pneumoniae, sensitive only to tetracycline.

This patient presented with frequent febrile illnesses which ultimately were demonstrated to be due to infectious agents. Patients with leukemic reticuloendotheliosis are very susceptible to infectious diseases (2). In attempting to

determine the causative agent in each case, it was not possible to rely on previously published reports, but appropriate cultures and tissue examination had to be performed each occasion fever developed. His course serves to re-emphasize a number of principles of infectious diseases:

1. Any person who is a compromised host who presents with fever must be assumed to have an infection. If initial cultures and laboratory tests are unrewarding, exhaustive efforts must be expended to determine the causative agent. If initial cultures are negative, one must reculture. If pulmonary infiltrates are present then the first step is to perform bronchoscopy with trans-bronchial biopsy and procede to lung biopsy if studies are negative (3, 4). Initially, a liver biopsy was not performed since liver function tests were not abnormal; however, in retrospect the liver biopsy might have provided an answer to the infection earlier. A bone marrow culture was negative early in infection although the bone marrow was shown to be the most sensitive site in previous reports (5, 6). Bacterial, AFB and fungal cultures as well as tissue examination must be performed on any suspicious lesion in the granulocytopenic patient with fever (7). One cannot rely on the usual physical findings of infection: fluctuation, swelling, exudation, local heat, or regional adenopathy, since these are less prominent in the granulocytopenic patient. Fever may be the only clue to infection. In this case, the eyelid lesion considered to be a staphylococcal infection, in fact represented an uncommon manifestation of disseminated histoplasmosis (5).

2. The potential infections represent such a wide variety with such variable sensitivity that empiric choice of antimicrobial agents is impossible. If an acute bacterial process is apparent, then empiric choice is possible. The preferred drug in persons with rapidly fatal infections, as leukemia, is carbenicillin in combination with gentamicin (8). Another consideration is to include a cephalosporin since carbenicillin is not effective against Klebsiella and one cannot rely upon gentamicin unreservedly to cover staphylococcal bacteremia. If pneumonia or urinary tract infection are present, one should consider Klebsiella as a good possibility and add a cephalosporin. Unfortunately, the hospital acquired Klebsiella maybe resistant to many antibiotics. Although amikacin, a soon to be released aminoglycosidic aminocyclitol, is effective against gentamicin-resistant klebsiella, it will not likely be effective in the granulocytopenic patient.

If a bacterial agent is not apparent by initial smears or within a few days by positive blood cultures, it becomes exceedingly difficult to predict the causative agent. The best approach is to attempt to determine the cause before launching across-the-board therapy. In this case, the disseminated granulomatous infections were slowly progressive. For one patient to have two disseminated granulomatous infections within one year is uncommonly bad luck. The subsequent appearance of another atypical mycobacteria in the blood adds a truly unique scale to this case. Mycobacteremia is extremely uncommon and in this instant was with an organism with limited sensitivities. The rapid-growing mycobacteria can be tested with the Kirby-Bauer method and the recommendation is to test all available antimicrobial and antituberculous agents, since one cannot predict which agent will be efficacious (Table 2). In this case, kanamycin was effective in vitro. The best therapy for all other atypical mycobacterial infections would include rifampin along with any other sensitive agent. Initial treatment of pulmonary disease due to M. kansasii, should be isoniazid and ethambutol. It is recommended that

TABLE 2

DRUG SENSITIVITIES OF ATYPICAL MYCOBACTERIA (SANDERS, C.V., PERSONAL COMMUNICATION)

Group I (# Tested)	INH (1 ug/ml)	Strep (10 ug/ml)	Rifampin	Etham-butal	Cyclo-serine	Vio-mycin	Ethio-namide
M. kansasii (89)	41	83	100	56	100	14	91
M. marinum (50)	0	84	96	96	100	46	88
Group II							
M. scrofulaceum (37)	8	43	17	50	91	21	38
Group III							
M. intracellulare (77)	3	10	6	54	73	5	8
M. terrae (21)	0	47	25	69	67	36	33
Group IV							
M. fortuitum (14)	0	21*	20	15	0	0	21
Unidentified (18)	0	12	25	31	13	13	6

* Kanamycin also = 20%

rifampin be reserved for re-treatment since INH plus ethambutol are equally efficacious in initial but rifampin is superior in retreatment of atypical mycobacterialpulmonary disease (9).

3. Not only are such patients susceptible to infectious agents, but they appear to be more prone to drug reactions. During the treatment of his infections, this patient manifested reactions to isoniazid, penicillin and rifampin. His reaction to isoniazid was rather atypical, but was eventually determined to be Sweet's syndrome by an alert dermatologist. Although he tolerated the other antituberculous therapy for a year, within 3 weeks of beginning amphotericin he developed a skin eruption. This was presumably due to rifampin since all the other drugs that he was on at that time were initiated later without recurrence of the rash. We hypothesize that the toxicity occurred because amphotericin increased the membrane permeability to the drug (10); in this case, presumably into epithelial cells.

Progressive disseminated histoplasmosis was the earliest form of the disease. Although it is not a commonly recognized entity considering the large endemic area for histoplasmosis, it continues to be a recognized cause of fever of unknown origin. It is a particularly lethal entity in children less than 1 year of age, who comprise up to 20% of the cases (5, 6). Males over 40 present with fever,with hepatomegaly, and oral mucous membrane lesions one half of the time. It is a rare cause of infection in the compromised host. It is only rarely detected in adult females with underlying diseases (Table 3). Serological studies are not very sensitive (Table 4) since only 1/3 to ½ have a positive skin test and a significant complement fixation titer of $\geq$ 1:32 is noted in only 1/3 of patients. The diagnosis of progressive histoplasmosis is made on examination of tissue for organisms. These are readily visible, even on H & E sections and can be confirmed by staining with the gomori methanamine-silver (GMS) stain. Impression smears of tissue on slides represent an excellent way to examine many more cells than possible with cut sections. These smears can be stained with Wright's or with GMS stain. Culture of tissue from mucous membrane lesions and lymph node frequently grow, but are only present in up to one-half (Table 5). Bone marrow culture has been the most sensitive test. Organisms will be cultured more often from liverbiopsy than organisms will be seen on tissue sections, which may require up to 6-8 weeks. The approach to a febrile individual who presents with hepatosplenomegaly or abnormal liver functions (Table 3) includes the following: liver biopsy with special stains for AFB and GMS, bone marrow culture with GMS stains of bone marrow biopsy material, sputum cultures which are obtained in the early A.M., early A.M. clean voided urine culture, and biopsy of any ulcerative lesion. Since histoplasma is particularly fastidious, any specimen must be submitted to the laboratory within 30 minutes. The importance of culturing for AFB and fungi is illustrated by another patient who presented to the VA shortly thereafter with fever, hepato-splenomegaly and leukopenia. He had worked at Fort Polk, Louisiana where he had been exposed to dust, following the bulldozing of barracks and other sites near trees where many birds, including grackles, had resided. The clinical diagnosis by the Infectious Disease Service was disseminated histoplasmosis, but liver biopsy demonstrated necrotizing granuloma. The culture subsequently grew Mycobacterium kansasii.

DISSEMINATED HISTOPLASMOSIS

TABLE 3

PRESENTING CHARACTERISTICS

	Rubin (5) %	Smith (6) %
Age < 1 yr	20	12
> 40 yr	48	65
Male	72	77
White	92	96
Fever	100	70
Hepatomegaly	68	62
Mucous membrane lesions	20	42

TABLE 4

SENSITIVITY OF IMMUNOLOGICAL STUDIES

	Rubin (5)	Smith (6)
Pos. skin test	56%	32%
C.F. Antibody (> 1:8).	61%	56%
(> 1:32)		36%

TABLE 5

SOURCE OF CULTURES	Rubin (5) % Positive	Smith (6) % Positive	Number Cultured (6) (26)
Oral lesions	-	91	11
Lymph node	-	72	7
Bone marrow	73	70	23
Sputum	43	60	20
Liver biopsy	-	57	7
Blood	27	54	24
CSF	-	45	11
Urine	-	43	23

B. Fever in compromised host with arterial involvement.

The 2nd case was a 69 year old white male who presented in August 1974 with GI bleeding, and who had to be explored because of a perforated small intestine. The pathological diagnosis was chronic lymphocytic leukemia. He was discharged on leukeran after an uncomplicated postoperative course. When he developed a Coomb's positive hemolytic anemia, he was treated with prednisone 80 mgs per day and cytoxan 100 mgs per day was substituted for leukeran. In June of 1975, he developed a high pitched, hoarse voice and on a clinic visit was noted to be thrombocytopenic (platelet count 69,000). On admission, he had dullness in the left base, a 3 component friction rub at the 3rd left intercostal space, and frequent PVCs. Admission chest x-ray showed cardiomegaly, but no pulmonary infiltrates. On the day of admission to the hospital, he spiked a temperature to 102^{o}F.His blood pressure dropped to 96/78 and an 8 mm paradoxical pulse was noted. An echocardiogram was negative for pericardial effusion, and central venous pressure was measured at 7 cms water. The patient was observed closely for tamponade. His platelet count rose to 144,000. Two days after admission, a left pleural effusion and widening of the mediastinum were noted. Pleural fluid revealed protein 2.6 gms%, WBCs 1500,(51% polys), and 600 RBCs. The mediastinal shadow & pleural effusion increased in size daily,so 4 days after admission an emergency aortogram was performed,showing no evidence for dissection. That evening the patient complained of weakness and inability to move the lower extremities. Eventually, a flaccid paraparesis localized to T 10 level. Cerebrospinal fluid exams revealed opening pressure of 12 cms of water, 70 red blood cells, white count to 186, with 72% polys and a protein of 80 mgs%. Smears for bacterial, AFB and India ink were negative. Irradiation was begun to the spine and mediastinum because of possible tumor infiltrates. The patient developed increasing respiratory difficulty, became hypotensive and died seven days after admission.

At autopsy,massively enlarged lymph nodes were noted in the mediastinum with a large, firm brownish yellow mass attached to the lingula of the left lung, and extending into the mediastinum and pericardium (latter had only 10 mls of clear fluid). The left pleural cavity contained dense, fibrous adhesions, with 650 mls of straw colored, clear fluid. The aorta was constricted just below the arch to a diameter of 1.5 cms by the mass but no evidence of dissection was noted. The spinal canal contained several areas of nodular material at T 12. Microscopic exam revealed broad, nonseptate hyphae which were invading blood vessels. The hyphae were in the anterior descending branch of the left coronary artery, (producing a coronary occlusion), mediastinal blood vessels and the spinal blood vessels (especially at T 12), Fungal cultures of the mediastinal tissue grew Mucor species. There was no evidence for chronic lymphocytic leukemia at autopsy.

This case illustrates a unique presentation of infection with Mucor species. The Mucor species, a member of the phycomycetes class, is recognized to cause rhinocerebral infections in diabetics with ketoacidosis and rarely produces pulmonary infarction in the diabetic patient (11). More aggressive chemotherapy

and prolongation of life of patients with leukemia and lymphoma has led to recognization of infections in leukopenic leukemic patients and in patients with lymphoma who have diabetes mellitus (12, 13). The hallmark of the infection is vascular invasion and infarction as in this case who presented with arterial widening simulating dissecting anuerysm and parapesis due to invasion of arteries leading to the spinal cord. The commonest site of infection has been the respiratory tract, characterized by the development of a pulmonary infarction, again due to the invasion of blood vessels by the hyphae (13). The clinical manifestations include new or changing infiltrates with cavitation or development of pleural effusion. In this case, the pleural effusion on the left side resulted from the mediastinal involvement, although it resembled the clinical presentation of left pleural effusion with dissecting aneurysm (14). Central nervous sytem involvement is also frequent,due to multiple blood vessel involvement leading to infarction and brain abscess. Cerebrospinal fluid usually reveals a small number of red cells, a moderate polymorphonuclear leukocyte response, elevated protein and a normal CSF sugar. Organisms are rarely demonstrated in secretions and are rarely grown prior to death. Hence, mucormycosis is to be considered in the patient with leukemia or diabetic with lymphoma who develops new or changing pulmonary infiltrates with fever, or clinical manifestations suggesting invasion of blood vessels. At autopsy, the patients frequently demonstrated other associated infections including bacterial and fungi (Aspergillus and Candida infections) and viral infections, particularly of the Herpes group (13).

C. Approach to compromised host with fever and pulmonary infiltrate.

Infections in the compromised host present a unique challenge to physicians in the 1970s. With an aggressive diagnostic approach, specific infections which are treatable can be recognized and specific therapy initiated. This is particularly applicable to the patient who presents with changing pulmonary infiltrates, in association with fever. If the platelet count is not low, nor any other bleeding abnormality present, a transtracheal aspirate should be performed to rule out acute bacterial infection. Many patients, in spite of low white count, develop pulmonary infiltrates(Case #1) and will demonstrate polymorphonuclear leukocytes in the sputum. The tracheal aspirate can be examined in the following manner (Table 6). If the patient does not have a bacterial infection which is

TABLE 6

EXAMINATION OF TRACHEAL OR BRONCHIAL SECRETIONS

OR TISSUE IN COMPROMISED HOST

Wet Mount: Fungal hyphae or yeast

Wright Stain: Histoplasma in monocytes

KOH preparation: Fungal hyphae (Blastomyces)

GMS Stain: Fungus or Pneumocystis

AFB and Gram Stain: Mycobacteria or bacteria

obvious by gram stain, then consideration should be given to a wide variety of treatable infections, including fungal,Pneumocystis and Toxoplasma infections (15, 16). Bronchoscopy with transbronchial biopsy should be performed immediately by pulmonary consultant. Certain institutions proceed immediately to a lung biopsy (4). However, we prefer the transbronchial route initially with the tissue submitted 1) in formalin to the pathology lab for both H & E stains and immediate special stains including GMS and 2) to the Mycology Lab for multiple smears and cultures. Impression smears can be made of the tissue (by physician if performed in emergency basis) and the smears then stained with Wright-Giemsa, GMS and both regular and weak (Fite) acid fast stains (for demonstrating Nocardia which is destroyed by the organic acid in the routine acid fast stain). If transbronchial biopsy fails to reveal a suitable explanation for the patient's pulmonary infiltrate, a lung biopsy can then be performed. The needle biopsy or aspirate appear to be satisfactory for demonstrating infection, although the open biopsy increases the success rate for underlying disease or non-specific inflammatory lesions (4), Table 7. The yield is greater for patients with lymphoma than for patients with other underlying disease. The value of a biopsy was real since the recovery rate for those with a treatable diagnosis was 70%,whereas it was only 25% if no specific diagnosis was made.

TABLE 7

DIAGNOSTIC YIELD BY PROCEDURE FOR PULMONARY DISEASE

IN COMPROMISED HOST (4)

Procedure	Underlying Disease	Non-specific Inflammation	Specific Infection	P.carinii	Fungi	Bacterial
			%			
Needle Aspirate	50	25	60	100	75	44
Needle Biopsy	50	66	75	100	-	-
Open Biopsy	97	100	79	100	83	50

Renal transplant patients who develop fever and pulmonary infiltrate are also likely to have a fungal infection (45% of autopsy cases) (17). The features of such patients are listed in Table 8. Since few had a positive culture in secretions; tissue examination would be in order. Unfortunately, they also had a high incidence of infection with other agents (17).

TABLE 8

FEATURES OF FUNGAL INFECTIONS

FOLLOWING RENAL TRANSPLANTATION (17)

	%
Male	96
Positive chest x-ray	90
Positive culture	43
Bacterial infection	61
Cytomegalovirus	57

D. Approach to patient with fungemia

The compromised host also has increased likelihood of developing fungemia, particularly with Candida species (18, 19). Candidemia occurs commonly (Table 9) in patients receiving antibiotics, those with intravenous catheters in place, particularly patients receiving parenteral hyperalimentation. Shock occasionally is noted (15% of cases). Fungemia with Candida may represent either transient catheter-induced fungemia, which is clinically insignificant,or disseminated fungal infection. Many patients will have transient fungemia which clears when the intravenous catheter is removed, antibiotics are discontinued and chemotherapy discontinued. The critical issue in these patients is to determine if tissue invasion has occurred. The most frequently involved organ systems at autopsy are the kidneys, myocardium, and less frequently the lung. The patients at greatest risk of having significant disseminated infections are those with fungemia who are neutropenic following immunosuppressive therapy (Table 10). One could predict an infection was present 89% of time if the patient with neutropenia following chemotherapy had a positive blood culture for Candida (19). Another clue to disseminated candidiasis was the isolation of Candida in cultures from at least three other sites, although this was not a sensitive test for the presence of candidiasis. However, if cultures were positive from three sites, this finding had a predictive value of 85% (Table 11). The approach in patients who are not immunosuppressed or leukopenic who have fungicemia would include: pull the intravenous catheter, discontinue antibiotics, and withhold specific therapy until post-catheter removal blood cultures are obtained (20). If these cultures are positive, then the patient should be treated. If the patient with fungicemia has hematopoetic or lymphoreticular malignancy or is leukopenic, particularly following immunosuppressive therapy, treatment should be initiated promptly (19). In addition, the fundus should be examined frequently. If a white, fluffy exudate is noted in chorioretina, then this is consistent with candidiasis and untreated is associated with a significant mortality (21, 22). This lesion was noted at autopsy in 85% of those having candidiasis (22). Another clue is the development of a maculonodular rash which shows pseudohyphae on microscopic examination and grows Candida (23).

TABLE 9

CLINICAL FEATURES OF PATIENTS WITH CANDIDEMIA (18)

	%
Antibiotics	91
Venous catheter	82
Surgical patient	56
Hyperalimentation	40
Shock	15

PROGNOSTIC FACTORS IN THOSE WITH FUNGEMIA:

TABLE 10

PRESENCE OF CANDIDIASIS IN THOSE WHO ARE NEUTROPENIC FOLLOWING CHEMOTHERAPY (19)

Candidiasis Present		Candidiasis Absent	
Neutropenia + Chemotherapy		Neutropenia + Chemotherapy	
Present	Absent	Present	Absent
25	4	3	8
Sensitivity:			86%
Specificity:			73%
Predictive Value			89%

TABLE 11

PRESENCE OF CANDIDIASIS IN RELATION

TO MULTIPLE POSITIVE CULTURES (19)

	Candidiasis Present		Candidiasis Absent	
Culture at 3 sites	Positive	Negative	Positive	Negative
	11	18	2	9
	Sensitivity:		38%	
	Specificity		82%	
	Predictive Value		85%	

Serological tests for Candida have also been developed to assess patients for presence of candidiasis. To evaluate these studies (Table 12), the sensitivity, specificity,and predictive value of test can be determined (24). The precipitin test for Candida has been very reliable when the patients have been highly selected for those with persistent fever, superficial candidal lesions (especially at IV catheter site) and in those with Candida endophthalmitis. In these situations, a positive precipitin test will indicate the disseminated infection 90% of the time (25). If cases are unselected as in individuals with prolonged hospitalization following cardiac surgery who have a low frequency of Candida infections, the test has low sensitivity and predictive value (26). The precipitin test in the Dallas area is performed by Dr. William Cooper, Director, Baylor Hospital Mycology Lab. He uses the counter-immunoelectrophoresis method, which gives an answer within 48 hours. A significant titer of 1:8 or greater is considered presumptive evidence for candidiasis (27). The only test available at the V.A. Hospital is the complement fixation test as a part of the fungal serology, but this test is a relatively insensitive test for candidiasis (Table 13).

TABLE 12

STATISTICAL ANALYSIS OF SEROLOGICAL TESTS

Disease Present		Disease Absent	
Test		Test	
True Positive (TP)	False Negative (FN)	False Positive (FP)	True Negative (TN)
Sensitivity = $\frac{TP}{TP + FN} \times 100$		The % of positive results in patients with disease	
Specificity = $\frac{TN}{TN + FP} \times 100$		The % of negative results among those who don't have the disease	
Predictive Value = $\frac{TP}{TP + FP} \times 100$		The % of positive results which are true positives	

TABLE 13

ANALYSIS OF PRECIPITIN TEST IN PATIENTS WITH CANDIDIASIS (25)

Candidiasis Present Test		Absent Test	
True Positive	False Negative	False Positive	True Negative
65	4	7	39
	Sensitivity	94%	
	Specificity	85%	
	Predictive Value	90%	

E. Prosthetic valvular endocarditis due to fungi

The 3rd case was a 63 y.o. BM with a long history of coronary artery disease, who had a homograft replacement of the aortic valve in 1972 for severe aortic insufficiency. The patient did well until October 1974, when symptoms of congestive heart failure ensued, requiring replacement of the aortic valve with a Starr-Edwards valve. Saphenous vein bypass grafts were also performed to the anterior descending and right coronary arteries. The patient spiked a temperature to 101°F on 1st post operative day, and increased sputum production was noted. Sputum cultures grew E. coli and he was treated with cephalothin. However, the patient continued to spike temperatures daily to 102 and began to complain of pain in the left shoulder. On the 6th post-op day, he developed transient, blurred vision on two occasions and had difficulty talking. No localizing neurological signs were detected on physical exam. He developed PVC's, and an EKG showed ischemia. The next day, he developed left bundle branch block with a first degree heart block. He continued to spike temperatures daily. On the ninth post-op day, he developed light-headiness, nausea, vomiting and a headache. Conjunctival petechiae were noted bilaterally, but no Roth spots were noted. Repeat sputum, urine and blood cultures were negative. On the tenth post-op day, he was noted to have hematuria. A brain scan was WNL, and an EEG showed bilateral temporal slowing. Later that day, the patient developed Cheyne-Stokes respirations, became more lethargic and developed a left hemiparesis. Since the patient was on Coumadin post-op because of the valve replacement, CSF examination was not performed. The patient developed a run of ventricular tachycardia, which responded to lidocaine, but he proceeded into coma. He developed a complete heart block, arrested and failed to respond to transvenous pacemaker and died on the 11th post-op day.

At autopsy, multiple small, dark purple, subendocardial foci were noted and a gray friable vegetation was present beneath the Starr-Edward valve, burrowing into, but not through the septum. Hemorrhage and necrosis were present at the margins of the vegetation. No vegetation was present on the valve, which appeared to be competent. Microscopic examination of the lesion revealed septate hyphae and cultures were positive for Aspergillus species. There were multiple microabscesses within the cerebrum, particularly around arterioles. These were presumed to represent septic emboli, although no fungi were identified in these lesions.

The causative agent in endocarditis which follows prosthetic valve surgery varies with the period of time in relation to the surgery (Table 14). The common causative organisms soon after surgery are: Staphylococcus epidermidis gram negative bacilli, fungi (Candida and Aspergillus) and Staphylococcus aureus (28, 29). In contrast, cases presenting late after surgery (greater than 60 days) have similar organisms as patients with subacute bacterial endocarditis (30) except that nonpathogenic coagulase negative staph (Staphylococcus epidermidis) is the most common staphylococcal organism (28, 29).

TABLE 14

CAUSATIVE AGENT IN ENDOCARDITIS

FOLLOWING PROSTHETIC VALVE SURGERY (28, 29)

	Early (≤ 60d)	Late (> 60d)	Usual causes of endocarditis (30)
	%	%	%
Streptococcus	11	49	60
Staph. aureus	18	15	18
Staph. epidermidis	27	19	2
Gram. Neg. Bacilli	20	9	1
Fungi	18	2	0
Miscellaneous	6	6	3
Negative Culture	0	0	16
Mortality	71	36	10

This case is fairly representative of those patients who develop Aspergillus endocarditis (31). It is most common after prosthetic valve surgery, is associated with fever and negative blood cultures,and the only diagnostic clue is a major arterial embolus, noted in 29% (Table 15). It is not apparent why blood cultures should be negative so consistently since Aspergillus sp. usually grow within 48 to 72 hours. One problem that might contribute to negative cultures is growth in an anaerobic environment. Any blood culture from patient with suspected fungal endocarditis should be referred to mycology laboratory for proper aerobic venting of bottle. No evaluation of serological test in Aspergillus endocarditis has been reported. Infection of the prosthetic valves with Aspergillus has been traced to contamination occurring during the operation or in the recovery room (32). Hence, contamination from the operating room air or pump equipment needs to be considered in any case that develops infection with fungi in this situation. Defects in ventilation system in the operating room needs to be identified and corrected if perchance an infection with Aspergillus develops. No further instances of Aspergillus endocarditis have occurred since November, 1974 at PMH. Proper therapy if the infection is recognized would require replacement of the valves (33). Hence, any clinical evidence of endocarditis soon after valve placement would suggest poor response and a need for valve replacement. Drug therapy alone does not appear to be successful but should be combined with reoperation (29).

TABLE 15

CLINICAL MANIFESTATIONS IN

PATIENTS WITH ASPERGILLUS SP. ENDOCARDITIS (31)

	%
Fever	87
Embolic Signs	70
Major Arterial Emboli	29
Petechiae	38
CNS Symptom	20
Pos. Blood Culture	8

Fungal endocarditis (Table 16) has been recognized in a number of clinical situations as well as a cause of prosthetic valve endocarditis (34). In addition to Aspergillus, Candida sp. (particularly Candida albicans) have been associated with infections of prosthetic valves. Ironically, most of the cases of Candida endocarditis following prolonged antibiotic usage have been in individuals treated for bacterial endocarditis (34). Rarely, the development of fungemia in a sick individual on prolonged hyperalimentation is associated with development of endocarditis (18). Again, the clinical clue is the development of major embolic phenomenon. In narcotic addicts who present with endocarditis (Table 17), up to 10% of the causative organisms are Candida species (35). The predominant Candida species is Candida parapsilosis. It is likely that those addicts who developed Candida endocarditis carry these organisms on their skin as they carry Staphylococcus aureus, the most common cause of endocarditis (36). Recently, Dr. Mackowiak has shown that heroin addicts carry Candida species in their oral mucosa more commonly than other individuals (37). However, the species was invariably Candida albicans. Hence, the explanation for the unique causative organism in addict endocarditis remains unclear. The treatment for any recognized fungal endocarditis is surgery (33). Patients fail to respond to chemotherapy alone, probably because the therapeutic agents penetrate poorly into fungal vegetations or experimentally into artificial blood clots (38). Ultimately, the best approach for fungal endocarditis is prevention. Measures should be taken to avoid introduction of fungi via either aerosolization in operating room or by proper care of intravenous catheters following surgery or during hyperalimentation (39, 40).

TABLE 16

CAUSATIVE ORGANISMS IN FUNGAL ENDOCARDITIS (34)

Condition	No.	Fungi	%
Cardiac surgery	82	Aspergillus	40
		C. albicans	37
		Candida sp.	23
Antibiotics + IV's	29	C. albicans	52
		Candida sp.	34
Heroin addicts	23	C. parapsilosis	52

TABLE 17

CAUSATIVE ORGANISMS IN INFECTIVE ENDOCARDITIS

IN NARCOTIC ADDICTS (35)

	Addicts (%)	Control (age matched) (%)
St. viridans	0	55
Other strep	14	14
Staphylococci	48	7
Gram-negative	24	10
Candida	10	0
Unknown	4	14

II. Pulmonary Cavitary Disease

A. Differential diagnosis of apical cavitary disease

The fourth case is a 43 year old white male from Fort Worth, Texas, who presented with 2-3 weeks of cough, anterior chest soreness and night sweats. He had been in good health until 5-6 months before admission when he noted 20 lb. weight loss, increased weakness, and increasing dyspnea on exertion. He had smoked hand-rolled cigarettes (Prince Albert) for 20 years. He had no hemoptysis and no productive sputum until 3 weeks before admission. His physical examination was within normal limits. Laboratory findings were unremarkable. His chest x-ray revealed bilateral apical cavitary disease. Multiple sputum smears for acid fast organisms were negative. Skin test with PPD was negative with a positive test with Candida antigen. Because the initial AFB smears were negative, bronchoscopy was performed. A slight amount of purulent secretion was noted in the apical posterior bronchus of the left upper lobe, but no mucosal abnormalities were noted. Transbronchial biopsy revealed inflammation but no granuloma. He was begun on isoniazid, ethambutol and penicillin V., 750 mgs qid. Approximately 4 weeks following bronchoscopy, a culture grew Histoplasma capsulatum. Fungal serologies showed Histoplasma yeast titer of 1:8 and the histoplasmin (mycelial) was negative. The patient was readmitted and Amphotericin B therapy was instituted with plans to treat with at least 2 grams.

This case illustrates the necessity for performing more extensive evaluation in individuals with pulmonary cavitary disease in whom a diagnosis of pulmonary tuberculosis cannot be made by smears or culture. There were no epidemiological clues in this case. Another patient seen at the DVAH at the same time grew Sporotrichum schenkii from tissue obtained at bronchoscopy. Bronchoscopy should be performed in the patient with apical cavitary disease when repeated smears for AFB are negative and if a PPD is less than 12 mm. A reaction of more than 12 mm to PPD is specific for M. tuberculosis but less than 12 mm is possibly due to atypical mycobacteria (41). Although a recent study has raised the issue that up to 17% of patients with active typical tuberculosis may have a negative reaction (42), better performed studies have found the negative reaction rate to be less than 5% (43). If the patient has a negative reaction, anergy must be considered: such as, immunosuppressive disease or medication, age (>70), leucocytosis ($>15,000$), and azotemia (44). If none of these are present and if a repeat PPD after 1 week in hospital is still non-reactive and smears are negative, then further workup with bronchoscopy is indicated.

Description of tests most often employed in Mycoserology (prepared by Dr. William Cooper, Director, Mycology Lab., Baylor University Medical Center, Dallas).

A. Aspergillosis
   1. Immunodiffusion test (Table 18) (45).
      a. Patients with allergic bronchopulmonary Aspergillosis usually have 1-2 precipitin bands by ID.

b. 3 or more precipitin bands suggests either invasive Aspergillosis or Aspergillus fungus ball. May be positive in patients with histoplasmosis.
c. Invasive disease in immunosuppressed patient rarely positive (46).

B. Blastomycosis (Table 20)
1. ID test is considered most sensitive test, but patients with histoplasmosis, coccidioidomycosis, and South American Blastomycosis may cross react.
2. Complement Fixation: sensitivity less than 50%. Not specific since cross-reacts in patients with acute histoplasmosis (47, 48).

C. Candidiasis (candidosis)
1. Counterimmunoelectrophoresis (CIEP)
a. Sensitive and quick test - results within 48 hours. Requires titration of antigen with each patient's serum followed by titration of serum with optimum antigen dilution.
b. Serum titers of 1:4 - 1:16 suggest systemic infection(27).
c. Serum titers of 1:2 - 1:4 suggest colonization by Candida.
d. This is a relatively new test that is quite promising but requires careful evaluation.
2. Immunodiffusion (ID)
a. Not as sensitive as CIEP and requires longer to carry out.
3. Complement Fixation - less sensitive on VA fungal serology.
4. Indirect Fluorescent Antibody, Latex Agglutination: Less specificity.
5. Whole Yeast Cell Agglutination
a. Can be used to quantitate results; fourfold rises or falls in titer have diagnostic significance.
b. Many normal people have titers of 1:160 or less

D. Coccidioidomycosis
1. Latex agglutination
a. (acute cocci) test: screening test
b. This test gives results comparable to the standard tube precipitin test; hence IgM antibody.
c. Sera from patients with acute pulmonary coccidioidomycosis would be expected to give a positive reaction with this test during the first 2-6 weeks of illness.
d. The test becomes negative approximately 6 weeks into illness, regardless of the patient's condition.
2. Immunodiffusion
a. Chronic cocci screening test.
b. Results of the ID test correlate very well with the complement fixation test, i.e. any patient with a positive ID test would be expected to have a positive CF titer. It is more difficult to quantitate the ID test than it is to quantitate the CF test.
3. Complement Fixation
a. The CF test has both diagnostic and prognostic value. "The higher the CF titer the more severe the disease" holds true for Coccidioidomycosis.
(1) A cocci patient who is getting well should have a declining CF titer.
(2) Exception: Patients with Cocci meningitis may have a relatively low CF titer in serum.

b. Patients with a positive CF titer in their CSF have either cocci meningitis or lesions close to the Central Nervous System. The serum should also be positive but perhaps at a lower titer, and the patient should also have symptoms of CNS disease.
c. Skin test with coccidioidin does not lead to rise in CF titer.

E. Cryptococcosis (49, 50)
1. Test for Cryptococcal Antigen (latex agglutination test with anti-cryptococcal globulin tagged latex particles).
a. The test can be performed on CSF, serum, or urine.
(1) CSF is the preferred specimen in patients with meningitis.
(2) Serum or urine are the preferred specimens in patients with widely disseminated disease.
b. Any positive titer is strongly suggestive of Cryptococcal disease (normal people do not have Cryptococcus antigen in their body fluids).
c. Titers are both diagnostic and prognostic. With successful therapy, the titer should decline over a period of several weeks.
2. Tests for Cryptococcal antibody
a. Whole cell agglutination, charcoal particle agglutination, and indirect FA tests have been described.
b. This test is most useful with patients who had mild pulmonary cryptococcosis, a single isolated Cryptococcoma or early, mild CNS disease. Tests for antibody run with CSF are almost always negative.

F. Histoplasmosis
1. Histoplasma latex agglutination test (51).
a. Positive in acute pulmonary histoplasmosis; positive or negative in disseminated disease; usually negative in chronic pulmonary disease.
b. Rising titer suggests dissemination of the disease.
c. High sensitivity but low specificity.
2. Immunodiffusion test (ID)
a. Especially useful with anticomplementary sera.
b. Presence of multiple bands (especially H & M) suggests current infection.
3. Complement fixation (51, 52)
a. Histoplasmin and yeast phase antigens employed (Table 19).
b. Titers of 1:32 or higher are strongly suggestive of histoplasmosis. Higher the titer, the more likely a sputum culture will be positive (6 or more).
c. With successful therapy titer should decline but may go back up to pretreatment level. A titer above the pretreatment level suggests relapse or dissemination of the disease.

G. Hypersensitivity Pneumonitis: "hypersensitivity pneumonitis precipitins."
1. Immunodiffusion test with a battery of antigens known to cause hypersensitivity pneumonitis.
2. The particular antigen to which any individual patient is sensitive may not be included in this battery. Such patients require extensive study and consultation with University of Wisconsin.

H. Paracoccidioidomycosis (South American Blastomycosis
   1. Sent through State Health Lab and must be accompanied by the patient's clinical history.
   2. Immunodiffusion and complement fixation tests have been described. The ID test gives the best results.
   3. Patients with South American Blastomycosis may cross react in serological tests for histoplasmosis and blastomycosis.

I. Phycomycete Infections
   1. Experimental at the present time.
   2. Contact Dr. Leo Kaufman who is the director of the fungus immunology unit at CDC for information.

J. Sporotrichosis
   1. Available at CDC only. Send request through State Health Lab in Austin and accompany request with a clinical history of the patient.
   2. Whole cell agglutination and latex particle agglutination tests appear to be the most reliable of those currently available at CDC.

K. Availability of mycoserology tests in Dallas are in Table 21.(on page 23).

ANALYSIS OF FUNGAL SEROLOGY

TABLE 18

PULMONARY ASPERGILLOSIS (45)

Test	Sensitivity %	Specificity	Predictive Value
Complement fixation	88	95	94
Immunodiffusion	80	99	98

TABLE 19

PULMONARY HISTOPLASMOSIS (CHICK)

Complement Fixation Titer

Infection	Present	Infection	Absent
$\geq$ 1:32	$<$ 1:32	$\geq$ 1:32	$<$ 1:32
16	11	51	200
Sensitivity		60%	
Specificity		80%	
Predictive Value		24%	

TABLE 20

PULMONARY BLASTOMYCOSIS (47)

Disease Present		Disease Absent	
Complement Fixation Titer ($\geq$ 1:8)			
TP	FN	FP	TN
3	6	2	6

Sensitivity	33%
Specificity	75%
Predictive Value	60%

BLASTOMYCOSIS (48)

	# Pos	# cases	Sensitivity (%)
Blasto	8	26	24
Histo	7	27	21

B. Mycetoma in pulmonary cavitary disease

The fifth case was a 57 year old white male, from Ennis, Texas, who presented with a 20 year history of chronic obstructive pulmonary disease. He had been treated in the East Texas State Hospital for atypical tuberculosis in the past. In November of 1973, he presented with a 6 month history of hemoptysis and x-ray revealed bilateral cystic disease, particularly in the upper lobes. Workup for active pulmonary infection at the time was completely negative. He presented for his final admission in December of 1974, with progressive dyspnea, productive cough and intermittent hemoptysis. He was in mild respiratory distress, with scattered rhonchi bilaterally. His pO_2 was 68 mm, his pCO_2 29 and a pH of 7.55. His chest x-ray at this time showed a pedunculated, soft tissue mass projecting into the posterior right upper lobe cavity. Bacterial culture was negative, but the fungus culture grew Aspergillus species. Fungal agglutinins showed histoplasma 1:8 and Aspergillus 1:8. He was given antibiotics with no response. While awaiting a needle biopsy, and aspiration of the cavity, the patient developed changes in behavior, became anorectic, refused liquids and food and then arrested. At autopsy he was shown to have slight pulmonary congestion with a large, 15 cm. cavity in the right upper lobe, which contained large nodules of amorphous necrotic tissue. Microscopically, this showed a mass of mycelium

TABLE 21

SEROLOGICAL TESTS FOR FUNGUS DISEASES

Disease	Type Test*	Availability†	Specimen	Request
Histoplasmosis	L.A.	P.M.H., B.M.L.	Serum	Acute Histo Test
	I.D.	T.S.H.L.	Serum	Histoplasma ID Test
	C.F.	T.S.H.L. V.A.R.L.	Serum	Fungal Serology
Blastomycosis	I.D.	T.S.H.L.	Serum	Blastomycosis ID Test
	C.F.	T.S.H.L. V.A.R.L	Serum	Fungal Serology
South American Blastomycosis (Paracoccioidomycosis)	I.D.	C.D.C.	Serum	I.D. Test for South American Blastomycosis
	C.F.	C.D.C.	Serum	CF for South American Blastomycosis
Coccidioidomycosis	L.A.	B.M.L.	Serum	Acute Cocci Test
	I.D.	B.M.L.	Serum	Chronic Cocci Test
	C.F.	T.S.H.L., V.A.R.L.	Serum,C.S.F.	Fungal Serology
Candidiasis	C.I.E.	B.M.L.	Serum	Candida precipitins
	C.F.	V.A.R.L.	Serum	Fungal Serology
Cryptococcosis	L.A: Antigen	B.M.L.,V.A.R.L. T.S.H.L.	Serum,C.S.F. Urine	Cryptococcus Antigen Test
	Antibody	V.A.R.L.,T.S.H.L.	Serum	Cryptococcus antibody
Aspergillosis	I.D.	B.M.L.	Serum	Aspergillus precipitins
	C.F.	V.A.R.L., T.S.H.L.	Serum	Fungal Serology
Hypersensitivity Pneumonitis	I.D.	B.M.L., C.D.C.	Serum	Hypersensitivity pneumonitis precipitins
Sporotrichosis	L.A.	C.D.C.	Serum	"Sporotrichosis" antibod

† P.M.H. - Parkland Memoral Hospital
B.M.L. - Baylor Mycology Laboratory
T.S.H.L. Texas State Health Laboratory, Austin, TX
C.D.C. - Center for Disease Control, Atlanta, GA
V.A.R.L. V.A. Reference Library.

* L.A. - Latex Agglutination
I.D. - Immunodiffusion
C.F. - Complement Fixation
C.I.E. - Counterimmunoelectrophoresis

(typical for Aspergillus) within an old tuberculous cavity. There was no apparent invasive disease outside the cystic cavity.

This patient presented with a rather typical picture for aspergilloma or invasion of a cavitary lesion with Aspergillus (53). As is commonly noted in patients who present with mycetoma, hemoptysis is more frequently present than in the usual person with cavitary disease (54). Many patients with obstructive pulmonary disease may grow Aspergillus in sputum cultures as a contaminant, but if this is found in association with the typical radiological picture, then this represents locally invasive aspergillosis. Serological studies for Aspergillus indicate that the complement fixation or immune diffusion test are sensitive, highly specific tests (Table 18), with a greater than 90% predictive value of revealing invasive pulmonary asperigillosis (45).However, patients who are compromised host who develop Aspergillus infection in association with chemotherapy for malignancies or hematopoietic diseases, do not develop antibody (46). In these patients, one must rely upon tissue diagnosis. The development of a new pulmonary infiltrate which cavitates in the absence of evidence for bacterial infection is highly suggestive of pulmonary aspergillosis. Recently, hospital-acquired aspergillosis has been noted in patients with underlying disease (55, 56). The sources of infection appeared to be 1) the ventilation system of an old hospital (55) and 2) fireproofing material in ceiling of a new hospital facility (56). Hence, if clusters of infections occur, a source within the hospital must be sought.

III. Other Miscellaneous Syndromes With Fungi

A. Rhinocerebral infection

The sixth case is a 59 year old black male, sewer inspector for the City of Dallas for 30 years, who presented with a 6 year history of painless swelling in the infraorbital region on the left side of the face. He was admitted initially in January of 1971 with mild swelling near the inner canthus of the left eye. Sinus films revealed a mass in the maxillary sinus, so a Caldwell-Luc procedure was performed. Pathological examination revealed granulomatous reaction and nonseptate hyphae consistent with Mucor were visualized. After surgery, the patient did well for about 6 months, but the swelling returned, and gradually increased over the next 5 years, such that he was barely able to open his left eye. It was not painful at any time and he had little difficulty with nasal secretions. He came to the VA after expressing some purulent, bloody secretions from one nodule under the left eye. Upon segmental resection, microscopic examination revealed granulomatous reaction and broad, branching nonseptate mycelial.

His fasting blood sugars and two hour postprandial sugars were within normal range. His white count was normal, with a normal differential. He failed to react to any antigen on skin testing, but further evaluation of T-lymphocyte function was not done. His IgG, IgA and IgM levels were normal. He had had no history of previous infection and had never been admitted to the hospital, save for the biopsy of the mass. He was treated with amphotericin B for a 6 week period at doses of 40 mgs three times a week. His mass decreased markedly in size and he

was able to open his left eye completely. Because of the clinical response and because his 6 weeks of sick leave were up, he was discharged to be followed in Infectious Disease Clinic after receiving 815 mgs of amphotericin B.

This patient represents a unique, clinical presentation of an infection with a member of the Phycomycetes class. The organism has not been properly speciated by CDC, but appears to be a soil organism, rather than the Rhizopus or Mucor species, the most common agent of rhinocerebral infection in diabetic patients (11). In patients with diabetic ketoacidosis it is a very rapidly progressing infection. It has also been noted in patients with renal disease and a few with severe gastroenteritis (11). Acidosis appears to be the principal underlying state enhancing susceptibility to these agents rather than hyperglycemia per se. The diagnosis is generally made by examination pathologically, since the organisms can be seen quite readily with H & E, although the extent of the infection is more prominently displayed with the GMS stain. Recently, infection has been noted (as described above) in individuals with leukemia receiving chemotherapy (13). It also has been a complication of patients with severe burns who develop a rapidly advancing infection with softening of the tissue and the usual proclivity for vascular invasion (57). Diagnosis is best made by prompt biopsy, since culture may require 2-5 days for the causative organism to be demonstrated. The treatment includes local wide debridement, although amputation has been necessary in a number of cases. The rhinocerebral infection responds to amphotericin B if recognized soon enough.

B. Mucous membrane infection (case used with the permission of Dr. Ralph Tompsett, Chief, Medical Education, Baylor University Medical Center, Dallas, Texas).

The seventh case was a 52 year old white man, who was admitted to the hospital because of a sore tongue. 3 months prior to admission, he noted the onset of malaise, a sore mouth and a sore tongue with an enlarging tender mass in the right anterior neck. He had had no fever, chills, night sweats, or dyspnea. Physical examination revealed a swollen,indurated ulcer with a raised border on the right lateral margin of the tongue. The tongue was generally enlarged. The patient was edentulous superiorly but had multiple caries inferiorly. He had a 3X5 cm slightly tender anterior cervical node on the right. Chest examination showed only increased AP diameter with no rales. Chest x-ray revealed multiple lesions in the left upper lobe with cavitation. Biopsy of the tongue lesion revealed granuloma formation and GMS stains showed spherical cells, some of which were empty, resembling an artifact, a few were budding and one demonstrated multiple small buds studding the entire surface of the spherule. Culture revealed typical organisms for Paracoccidioides brasiliensis. Epidemiological history revealed the patient traveled for 20 years in the U.S. Navy and at various times was stationed in Puerto Rica, Trinidad and the British West Indies; however, he had never visited Brazil. Generally, he had only been in the port cities and never traveled into the interior of the continent.

The differential diagnosis for ulcerative lesions of the tongue and mucous membranes includes carcinoma, tuberculosis, syphilis, histoplasmosis and (in those with epidemiological history of having visited South America) paracoccidioidomycosis, or South American blastomycosis. The infection occurs predominantly in males over 30 who are principally farmers. Brazil is the principal site of infection, although other areas in South and Central America up to Northern Mexico have been shown to develop the infection. It has a characteristic predisposition for the mucous membranes and pulmonary distribution (Table 22), although rarely skin lesions will be noted (58, 59). Lymphadenopathy is noted in a large proportion of the cases, particularly cervical adenopathy. Biopsy will prove the diagnosis in most of the cases, although special stains (GMS) are required to reveal the organisms. Histoplasmosis is the principle fungi producing mucous membrane involvement in the United States and it primarily presents in males over 40 who also have pulmonary involvement (6). Again, biopsy is essential to reveal the diagnosis. It is important to remember that granulomas may not be seen, but Histoplasma organisms will be noted within monocytes in the inflammatory reaction. Skin tests and serological tests are available for Paracocidioidomycosis (Table 21). Although complement fixation studies are positive in 90% of the cases, there is a significant cross reaction with Histoplasma capsulatum.

TABLE 22

PRESENTING COMPLAINTS

PARACOCCIDIOIDOMYCOSIS

Complaint	%
Oral mucosal ulcer	52
Respiratory symptoms:	
Productive cough	37
Skin ulcer	10

C. Cutaneous infection

The following humorously written description of an epidemic of sporotrichosis is presented: (60)

"Thus, we were confronted with six patients with proven sporotrichosis. All lesions had been present for approximately two months, with onset about December 1. The data suggested a common source outbreak, curiously limited to members of a freshman class.

"The six patients were confronted as a group. They denied traumatic contact to known potential vehicles of sporotrichosis (rose bushes, cactus, salt meadow hay, barberry, carnations, moss, timber splinters, or other plant thorns). The only possible clue to the source was the mention of bricks by two of the patients. After considerable cajoling and chiding of the group, the following course of events was uncovered. The six were among several dozen freshmen recruited to transport bricks from a vacant lot to the site of construction of a new fraternity house. Remuneration was small, but the sun shone brightly on a warm Saturday morning in mid-November. A supply of beer was chilled and easily accessible nearby. The group decided to traverse the short distance by passing the bricks one by one, bucket-brigade style, from the lot to the construction site. As the ambient temperature rose and work progressed, inordinantly large quantities of beer were apparently consumed. As fate would have it, patient 4 dropped a brick on the bare toe of patient 2. Patient 2, in turn, hurled a brick at patient 4 - missing him - but striking patient 3 in the neck. Whereupon a melee ensued. Patient 5's aim at patient 6 was more accurate. The battle subsided as injuries mounted and the supply of malt beverage was exhausted. Each patient then readily admitted that lesions had developed at the site of brick-trauma.

"Following the epidemiological method, an attempt was made to find the bricks for culture. By this time, they were securely mortared in the west wall of the new fraternity house. Undismayed, a medical student was dispatched, chisel and hammer in hand, to retrieve a portion of the suspected vehicles of infection. Pieces of brick were diligently chiseled out and attempts were made to homogenize them for culture. This resulted in total destruction of the blades of our blendor, which was no longer under warranty. However, samples of a crude mixture of Spanish moss, soil, and residual packing straw, obtained from the site where the bricks had been stored for two years, yielded luxuriant growth of S. schenckii on culture. Unfortunately, the source of the packing straw and bricks could not be traced" (60).

Differential diagnosis of sporotrichoid-like lesion is given in Table 23. One must consider a number of infectious organisms when a person presents with a chronic picture of cutaneous and lymphatic involvement. If the patient presents with chronic draining ulcers with lymphangitic involvement and is a nursery worker or an alcoholic rose gardener (61, 62), it is likely that the diagnosis is sporotrichosis. The organisms are rarely demonstrated in secretions, but a culture will grow quite rapidly (3 - 7 days) at room temperature. If the person has a history of swimming in fresh water, particularly unchlorinated pools, or of working with a tropical fish aquarium and has traumatized the arms which leads to a lesion with abscess formation, then infection with Mycobacterium marinum is likely (63). Organisms may be demonstrated in tissue and cultures will be positive in 2-3 weeks, if grown at 25-30° C rather than 37° used for typical mycobacterium. If the person is from the southern United States and has a history of walking barefooted in the soil, then mycetoma must be considered (64). The typical mycetoma presents with chronic abscesses in which white to yellow granules, containing acid-fast granules if Nocardia or mycelia if a fungus is present. In the United States and in Central and South America, the causative agent is principally Nocardia brasiliensis although other species as Streptomyces madurae, or Allescheria boydii, may be the causative agent (65). The disease occurs primarily in males, who come in contact with soil, particularly if the feet are covered. Women working in a similar situation are protected from the disease. The Nocardia can best be demonstrated

TABLE 23

DIFFERENTIAL DIAGNOSIS OF SPOROTRICHOSIS-LIKE INFECTIONS

	Source of Infection	Clinical Features	Histopathological Findings	Culture
Sporothrix (Sporotrichium) schenckii (61)	Rose thorns, peat moss, hay; usually by direct injury from thorns, splinters or inoculation through open wound.	Chancre at site of inoculation followed by lesions along the lymphatics draining the area. The regional lymph nodes are enlarged.	Granulomatous reaction with suppuration. Organisms are rarely found.	Grows rapidly at room temperature as moist, or fluffy, creamy colonies.
Mycobacterium marinum (63)	Fresh water, especially swimming pools, tropical fish aquariums, watering troughs, etc. Infection occurs after direct injury or through a cut or abrasion.	Initial chancriform lesion with subsequent nodular abscesses along lymphatics. Regional lymph nodes are usually not enlarged.	Granulomatous reaction with or without tubercle formation. Organisms when present are acid-fast, plump, long and prominently beaded.	Grows on media for culture of mycobacteri in 2-3 weeks at 25-30° C, nitrate, negative; Arylsulfatase, negative 3d, positive 14d.
Pasteurella tularensis	Ticks and other arthropod bites: rodent bites.	Initial chancriform lesions with marked enlargement of the regional lymph nodes ("ulceroglandular tularemia").	Nonspecific granulomatous reaction with areas of focal necrosis.	Grows on blood agar in 6-10 days at 37 C as a pleomorphic gram-negative organism.
Nocardia brasiliensis (64)	Soil organism which enters skin by direct inoculation; male farmers in tropical zones who go barefooted.	Slowly developing, draining abscesses, usually multiple. Lower extremities; feet, ankles, but may be anywhere on skin: bones often affected.	Granulomatous tissue reaction with abscess formation with white or yellow acid-fast granules in pus. Tuberculoid granuloma is occasionally seen.	Grows slowly on Sabouraud's agar without antibiotics, as yellow chalky, moist colonies.
Blastomyces dermatitidis (48)	By dissemination from lungs; rarely by direct inoculation of skin.	Slowly growing local lesion with characteristic verrucous, elevated borders. Central scarring, with lymphadenitis, lymphangitis and nodules along course of lymphatics.	Dense granulomatous infiltrate with many PMNs* and numerous single budding cells which are PAS positive.	Grows slowly 3-4 weeks as cream-colored colonies at 37 C.

* PMN = polymorphonuclear neutrophil leukocyte.

by using a weak acid fast stain. Granules may be seen,resembling sulfur granules typical for Actinomyces, since draining cutaneous lesions with Nocardia may be associated with granules (66). Blastomycosis must be considered if cutaneous lesions have a verrucous border, or more typically, have a central ulcer with elevated borders (48). Lymphadenitis and lymphangitis may be noted. GMS stain of tissue demonstrate the organisms, although H & E preparation may show typical organisms. The cultures tend to require 2-3 weeks, but the diagnosis can usually be proven by smear.

Sporotrichosis also may present as extracutaneous infection (67), specifically as tenocynosynovitis (68), osseous or arthritis (69), or as pulmonary disease (70), in which it resembles chronic cavitary disease in person with underlying lung disease.

Table 24 gives the mycology and Table 25 the summary of clinical features of fungus infections.

TABLE 24

MYCOLOGY OF FUNGUS INFECTIONS

	Smears		Culture	
	Secretions	Tissue	Length (Wks)	Characteristic
Coccidioidomycosis	Wet mount (NaOH)	H&E GMS	2-3	Spherules-smear Arthrospore-culture
Histoplasmosis	Wright's Stain	H&E GMS	4-8	Tuberculated macroconidium
Blastomycosis	KOH prep	H&E GMS	4-8	Budding yeast at 37° C
Aspergillosis	KOH prep	GMS	1	Septate hyphae
Cryptococcosis	India Ink	Mucicarmine or GMS	1-4	Budding yeast at 37° C
Sporotrichosis	PAS	GMS	1-2	Conidia as bouquet
Mucor	PAS	H&E	1	Branching non-septate hyphae
Paracoccidioidomycosis (South American Blastomycosis)	GMS	GMS	2-3	Pilot's wheel cells

TABLE 25

SUMMARY OF EPIDEMIOLOGY AND CLINICAL FEATURES OF FUNGAL INFECTIONS

				Clinical Illness	
Organism	Method of Inoculation	Georgraphic Area	Groups Predisposed	Typical Presentation	Disseminated Form
Coccidioido-mycosis	Inhalation of dust	S.W. (lower U.S. Sonora Zone)	New inhabitants. Black more susc-eptible to dis-seminated.	"Flu"-like, arthralgias Erythema nodosum	Skin, bones, meninges (71)
Histoplasmosis	Inhalation: Bird droppings; Bat guano at cave entrance.	Middle U.S.: River Valleys (Miss., Ohio, Potomac).	All inhabitants, Spelunkers. Females: erythema nodosum	"Flu-like illness. Interstitial pneu-monia	Liver, spleen, bone marrow, kidney, adrenal. (6)
Blastomycosis	Inhalation ? Soil	Middle South	Construction, forest workers, farmers.	Cavitary disease.	Skin, prostate, bone (48)
Aspergillosis	Inhalation (vegetation)	Ubiquitous	Penguins, pigeon-crammers, patients with cavitary disease.	New growth in cavity	Lung, (54) endocardium (31)
Cryptococcosis	Inhalation Pigeon droppings	Urban	Pigeon fanciers, diabetes, Hodgkin's disease.	Pulmonary nodule	CNS, kidney (72)
Sporotrichosis	Contact & wound Inhalation	Ubiquitous	Rose gardeners, Forestry workers	Ulcer on skin, (61) cavitary disease (70)	Bone, joint (67) (69)
Mucormycosis	Direct invasion, Upper respiratory tract	Ubiquitous	Diabetic, keto-acidosis. Mal-nutrition. Leukop-enic leukemia	Rhino-cerebral, (11) orbital	Lung infarct (13) Arterial Invasion
Paracoccidioido-mycosis	Inhale spores	Central and South America	Farmers	Mouth ulcer,(58,59) Pulmonary infiltrate	Mucocutaneous

IV. Therapy

A. Amphotericin B.

Amphotericin B remains the treatment of choice for most fungal infections (73-75). It has clearly been established to improve the course of the disease and prevent death in progressive disseminated histoplasmosis (6, 74), histoplasmosis, blastomycosis (Table 26). In each of these diseases, the recommended quantity of amphotericin is at least a total dosage of 2 gms, since quantities less than that are associated with a significant relapse rate. Many of these patients have significant underlying lung disease and a shortened five year survival without therapy, but therapy is efficacious. After therapy, interpretation of new infiltrates represents a problem. A helpful clue to the activity of histoplasmosis is whether the complement fixation titer falls after therapy. If the titer were significantly elevated (greater than 1:32) before treatment, if the titer falls to zero and remains zero then the pulmonary problem is not activity of fungus but another problem. If the titer exceeds the previous titer, then it represents relapse (Chick, E.W.). One must not only evaluate the serological response but also the clinical condition and obtain appropriate cultures. Rarely does a person with chronic cavitary histoplasmosis develop disseminated infection. The more likely causative agent if the patient presents with hemoptysis and infiltrate within a cavity is that this represents aspergillosis (54). Surgery plus amphotericin B is the treatment for selected cases (74). Blastomycosis tends to relapse frequently, but treatment of moderate cases with either diamidine (2-hydroxy-stilbamidine), or amphotericin is associated with significant improvement of both pulmonary and cutaneous lesions (74). Cutaneous sporotrichosis is treated successfully with iodides, beginning with 1 ml of aqueous solution of potassium iodide(1 gram per milliliter) 3 times a day, and increasing every other dose by a ½ ml, until the dose reaches 4 ml :3 to 4 times per day for at least 4 weeks. Patients who do not respond to therapy or those patients who have osseous, pulmonary or disseminated sporotrichosis should be treated with amphotericin B (Table 27). Good success rates have been noted in patients with joint involvement,although surgery may be required in addition to amphotericin B. Amphotericin B is the treatment of choice for coccidioidomycosis. Major emphasis should be placed on early recognition of dissemination (71, 74) (Table 28).

Significant improvement in the course of cryptococcal meningitis has been noted following treatment with amphotericin B (72). Although up to 90% of untreated patients will die within a year, the overall success rate with amphotericin B approaches 80% (74). The significant risk factors for a poor response in cryptococcal meningitis are listed in Table 29 (76). Intrathecal therapy with injection of amphotericin B into an Ommaya reservoir is reserved for those patients who remain culture positive after 3 gms of amphotericin or for those with overwhelming infection (77). A good success rate has been reported with a 6 weeks course of the combination of amphotericin B (20 mgs daily) and 5-fluorocytosine orally (150 mgm per kg). However, hematologic complications developed in 9 of the patients, although renal insufficiency developed to only a mild degree in 6 patients (78).

TABLE 26

RESPONSE TO THERAPY

Therapy	% Improved	% Relapsed	% Died
Progressive Disseminated Histoplasmosis (74)			
None	8	0	92
Amphotericin (<10 mg/Kg)	10	-	90
Amphotericin (>25 mg/Kg)	88	9	30
Chronic Pulmonary Histoplasmosis (74)			
Therapy	% Improved	% Relapsed	% Died
None	32	-	50
Amphotericin	57	15	28
Amphotericin <2 gm	0	27	36
Amphotericin >2 gm	71	14	14
Blastomycosis (74)			
Therapy	% Improved	% Worsened	% Died
None	57	10	33
Diamidine	72	18	10
Amphotericin	79	9	11

TABLE 27

TREATMENT OF EXTRACUTANEOUS SPOROTRICHOSIS (74)

	Good Response %		
	Osseous	Pulmonary	Disseminated
Iodides	35	46	39
Amphotericin B	80	75	76

TABLE 28

CLINICAL EVIDENCE FOR DISSEMINATION

IN COCCIDIOIDOMYCOSIS (17)

Black male, pregnant female

Persistent fever at 4 weeks

New pulmonary infiltrate

Paratracheal adenopathy

Draining sinus (bone or lymph node)

Hepatomegaly, abnormal liver function

TABLE 29

SIGNIFICANT RISK FACTORS IN

CRYPTOCOCCAL MENINGITIS (76)

		Predictive Value (%)
	Cases	Death
Continued Steroid Therapy	3	100
No change in CSF Ag Titer	23	80
Positive culture > 2 sites	36	77
Initial CSF Ag ≥ 1:32	40	62
CSF ≥ 1:8, End of Treatment	22	40

B. Combination therapy

Significant synergism of amphotericin B and 5-fluorocytosine has been documented in a number of experimental studies (10). The mechanism for the synergism relates to the effect of amphotericin B on membranes of the fungi, such that these agents are able to penetrate the cell and inhibit growth. However, the increased penetration also may apply to mammalian cells since toxicity of the second agent may be enhanced (10, 78). A number of multicenter studies are being performed; amphotericin B and 5-fluorocytosine for cryptococcal meningitis, amphotericin B and rifampin for histoplasmosis, and amphotericin B plus polymyxin B for coccidioidomycosis. In the meantime the only indication for the combination of 5-fluorocytosine plus amphotericin B is in individuals with disseminated candidiasis and very ill individuals with cryptococcal meningitis, as those with Hodgkin's lymphoma or leukemia. In the usual case of cryptococcal meningitis, the combination probably should be reserved for a multicenter protocol.

Admonitions on therapy with amphotericin B (for more detail see references 73 and 79).

1. Amphotericin B must be diluted only in 5% dextrose in water. Addition of saline will aggregate this colloid.
2. Amphotericin B is stable if infused the same day, although unused portions must be refrigerated and protected from light. Infusion bottles do not have to be covered with a brown bag, no matter what charge nurse says.
3. The suspension should be administered over a 4-6 hour period. The first treatment should be with a small dose (5 mg) and then the dose can be increased 10 mg daily until 40 to 50 mg is reached and renal function stabilized. Tolerance to toxic effects does develop. Don't let interval between administration exceed 2-3 days, or toxic reactions (fever, nausea, etc.) will occur. Each patient develops own idiosyncrasies of administration, so let patient determine rate and length of infusion.
4. Follow renal function and potassium carefully. Decrease dose or hold dose same if serum creatinine exceeds 2.5 mg%. Although the creatinine clearance after treatment never quite reaches pre-treatment value, it approaches normal. Certainly, no progressive renal disease occurs (although calcifications may be seen in kidneys if biopsied). Renal toxicity is magnified once total dosage exceeds 4 gms.
5. Each patient should have flow sheet with following data:

TABLE 30 (79)

RECOMMENDED AMPHOTERICIN CHART

Date	Amphotericin B		Bun/Cr	Hb/Hct	K+	Cerebrospinal Fluid (If Meningitis)			Misc
	Daily	Total				Cells	Sugar/Protein	Culture	

6. The dose of amphotericin B does not need to be altered in patients with renal insufficiency, nor during renal dialysis (80). The agent appears to be highly protein-bound and is poorly dialyzable. In contrast, the dosage of fluorocytosine does need to be reduced in renal failure. One recommended schedule (73) is to use one dose q 6 hours for creatinine clearance = 40, q 12 hrs for 20-40, and q 24 hours for 10-20 ml/min. If the patient is being dialyzed, a recommended schedule is to give 25 to 50 mg/Kg immediately after hemodialysis every 48-72 hours (80). It is preferable to measure blood levels, but these are not available presently in the Dallas area, although Dr. Cooper at Baylor University Medical Center is planning to set up method.

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